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BRIEF REPORT

Atropine Disrupts Passive Avoidance Learning in Young Chicks[1]

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Four-day-old Vantress × Arbor Acre chicks were first trained to key-peck for heat reinforcement and then tested for passive avoidance learning following an intraperitoneal injection of atropine sulfate or saline. Chicks injected with 1.0 mg/kg of atropine responded more quickly than saline-injected chicks when their key-peck responses were punished with aversive wing-shocks. These findings, therefore, are consistent with the view that cholinergic mechanisms are involved in inhibitory behavior in the young domestic chick.

Scopolamine, a cholinergic antagonist, has recently been shown to affect the young domestic chicks' performance on three tasks that have been assumed to measure inhibitory behavior (Zolman, Mattingly, & Sahley, 1978). Specifically, 4-day-old chicks injected with scopolamine were found to be more active in an open field, more resistant to extinction after key-peck conditioning, and disrupted in key-peck passive avoidance (PA) learning when compared to saline-injected control chicks. These scopolamine effects on the young chicks' behavior are similar to the reported effects of scopolamine in rats (see Bignami, 1976) and suggest that for the precocial chick, like the altricial rat, response suppression may be mediated by cholinergic mechanisms.

Scopolamine also produces a decrease in the duration of tonic immobility in chicks (Hicks, 1976). However, atropine, another cholinergic antagonist, apparently does not affect the duration of tonic immobility in chickens (Hicks, 1976; Maser, Gallup, & Hicks, 1975). As both scopolamine and atropine are antimuscarinics, Ksir (1979) has proposed that the decrease in tonic immobility duration produced by scopolamine may be unrelated to its anticholinergic activity.

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Since the effects of atropine on other inhibitory behaviors of the young chick have not been studied, the purpose of the present study was to determine whether atropine, like scopolamine, would disrupt the PA learning of the young chick. Therefore, separate groups of 4-day-old chicks were first trained to key-peck for heat reward (autoshaping and acquisition training), injected with various doses of atropine or saline, and then their subsequent key-peck responses were punished by aversive wing shocks (PA testing).

Forty-eight Vantress × Arbor Acre chicks were incubated and hatched at 37–38°C and 56–60% relative humidity. All chicks were reared in groups of 10–12 in white Plexiglas brooder compartments (56 × 33 × 23 cm) in a 35°C room with food and water available ad lib. Behavioral testing was done in four conditioning boxes housed individually in 10°C incubators. Chicks were first trained to peck an illuminated key (a white, 3.2 × 2.2-cm bar presented vertically on a red background) to receive warm 35°C air delivered through the bottom of the conditioning box. On autoshaping trials heat reward was scheduled to occur automatically at the end of each trial (key-light offset), but was given immediately when the chick pecked the illuminated key during the trial. This autoshaping procedure trained the chick to peck quickly when the key was illuminated. On acquisition and PA trials, however, the chick had to peck the illuminated key to receive heat reward; that is, reinforcement was not scheduled to occur at the end of the trial. Finally, on PA test trials each key-peck response was also punished by a 5 mA–.5 sec wing shock (see Mattingly & Zolman, 1980, for a detailed description of the conditioning boxes, the heat reward system, and the wing-shock procedure).

Behavioral testing consisted of two autoshape sessions, an acquisition-PA session, and then three PA sessions. Each session consisted of 24 discrete trials separated by a 20-min intersession interval. One hour before training the chicks were isolated in individual white Plexiglas cylinders (20 × 15 cm), and were returned to these isolation cylinders after each training session.

The scheduled sequence of events on a trial during autoshaping was: (1) key-light on for 16 sec; (2) key-light offset with 8 sec reinforcement (35° air); (3) 5-sec intertrial (ITI) with house light on; (4) key-light onset, etc. If the chick pecked the key during the 16-sec stimulus duration, however, reinforcement was delivered immediately, and a new trial started after the 5-sec ITI. Immediately following the first autoshape session, all chicks were wing-clipped for shock delivery and were then run in their shock harness during all of the following sessions. After the second autoshape session, chicks that made 12 or more responses were weighed, and then injected intraperitoneally with either isotonic saline or atropine. Four different groups of 12 chicks each were given either 0 (saline), 0.5, 1.0, or 2.0 mg/kg of atropine sulfate as the active base. All doses were given in a

volume equal to 1% of body weight, and drug conditions were coded so that group assignments were unknown to the experimenter during injection and testing procedures. The acquisition-PA session began about 20 min after drug injections.

Mean response latencies across blocks of 12 trials for the groups on the two autoshape, the single acquisition-PA, and the three PA sessions are presented in Fig. 1. The chicks significantly decreased their response latencies from the first to second autoshape session, $F(1, 44) = 73.60, p < .0001$. Also, the chicks responded more quickly on the second block of 12 trials of each session than on the first block, $F(1, 44) = 32.61, p < .0001$. As expected, there were no significant differences among the chicks in the four groups during the two autoshape sessions before drug injections were given.

Since the acquisition-PA session included 12 acquisition and 12 punishment trials, this third session was analyzed separately from the subsequent three PA sessions which included all punishment trials. On this session chicks in all groups showed a significant increase in their response latencies when punishment conditions were introduced—block effect, $F(1, 44) = 61.64, p < .0001$. Although the atropine-injected chicks had higher response latencies than the saline-injected chicks on the first block of 12 acquisition trials, this difference was not significant.

The chicks continued to increase their response latencies across the three subsequent PA sessions, $F(2, 88) = 8.06, p < .001$, but this increase was greater for saline- than for atropine-injected chicks—Group × Session interaction, $F(6, 88) = 2.62, p < .05$. Analysis of this significant Group × Session interaction indicated that the 1.0 mg/kg atropine chicks

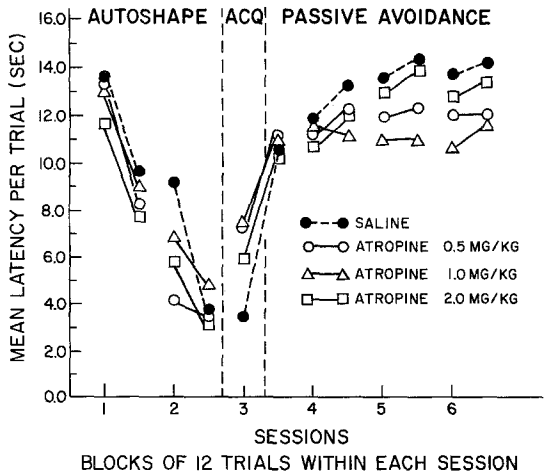

FIG. 1. Mean response latencies per trial across blocks of 12 trials for the saline and atropine groups of chicks during two autoshape, one acquisition-PA, and three PA sessions.

responded significantly more quickly than the saline-injected chicks on the last two PA sessions—Newman–Keuls tests, $p < .05$ in each case. Also, the response latencies of the 0.5 and 1.0 mg/kg atropine groups did not significantly increase across the last three PA sessions, $p > .05$ in each case. The 0.5 and 2.0 mg/kg atropine groups' response latencies, however, did not significantly differ from those of either the saline or the 1.0 mg/kg atropine groups.

The disruption of PA learning produced by 1.0 mg/kg atropine in the present study is similar to that produced by scopolamine (Zolman et al., 1978), and indicates that the retardation of PA learning in young chicks produced by these cholinergic antagonists is probably related to their receptor blocking activity rather than to nonspecific pharmacological effects (see Ksir, 1979). In most behavioral tests, cholinergic agonists and a quaternary cholinergic antagonist can be used to demonstrate the pharmacological specificity for the effects of cholinergic antagonists on the animal's performance. But the predicted effect of cholinergic agonists on the young chick's PA learning would be increased response suppression. A drug-induced increase in response suppression in a PA test is difficult to interpret because the drug could be interfering with motor capabilities. Indeed in the present study, the key-peck performance of chicks injected with 2.0 mg/kg of atropine was probably disrupted, and consequently, their response suppression on PA test trials was not different from that of saline-injected chicks, who stopped responding very quickly. (Of course, the advantage of using a PA learning test in pharmacological research is that a learning deficit is indicated by continued responding, and toxic or debilitating effects of a drug cannot be used to explain this deficit.) Also, since the blood–brain barrier is not fully functional in chickens until about 1 month after hatching (Spooner & Winters, 1965), the quaternary derivative methylatropine cannot be used to distinguish between central and peripheral effects of atropine on the behavior of 4-day-old chicks.

Cholinergic antagonists, besides affecting PA learning, affect several other inhibitory behaviors of young chicks. For example, scopolamine significantly increases both spontaneous locomotor activity and resistance to extinction of 4-day-old chicks (Zolman et al., 1978). Furthermore, atropine reduces significantly spontaneous alternation of 10-day-old chicks (Brown, 1976). Taken together, these results are consistent with the view that cholinergic mechanisms mediate inhibitory behavior in the chick. It should be emphasized, however, that most response suppression tests used in psychopharmacological research cannot differentiate among inhibitory, memory, or discriminative processes. Consequently, it cannot be concluded from such tests that cholinergic antagonists affect only inhibitory processes (see Zolman et al., 1978, for a review).

These general problems of interpretation, however, should not detract

from the finding that both scopolamine and atropine retard PA learning of the 4-day-old chick. Indeed, these data extend previous findings on the effects of cholinergic antagonists on PA learning in the young atricial rat (Blozovski, Cudennec, & Garrigou, 1977; Wilson & Riccio, 1976) to the 4-day-old precocial chick. Moreover, since atropine does not significantly affect tonic immobility in chicks (Hicks, 1976), the present results suggest that response suppression observed in PA learning and tonic immobility tests may be mediated by different neurochemical processes.

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